

P-ISSN: 2706-7483
E-ISSN: 2706-7491
NAAS Rating (2026): 4.5
IJGGE 2026; 8(5): 95-102
www.geojournal.net
Received: 05-03-2026
Accepted: 07-04-2026

Madhu Guguloth
ME Mining Engineering,
Department of Mining
Engineering, College of
Engineering, Osmania
University, Telangana, India

Sathish Kumar Mittapally
Professor, Department of
Mining Engineering, College of
Engineering, Osmania
University, Telangana, India

Ajay Eru
ME Mining Engineering,
Department of Mining
Engineering, College of
Engineering, Osmania
University, Telangana, India

Srinivas Kuchibhotla
Professor, Department of
Mining Engineering, College of
Engineering, Osmania
University, Telangana, India

Corresponding Author:
Madhu Guguloth
ME Mining Engineering,
Department of Mining
Engineering, College of
Engineering, Osmania
University, Telangana, India

Integrated application of agriculture waste Biochar and *Rhizobium* for the restoration of acidic mine overburden soil: A biogeochemical approach

Madhu Guguloth, Sathish Kumar Mittapally, Ajay Eru and Srinivas Kuchibhotla

DOI: <https://www.doi.org/10.22271/27067483.2026.v8.i5b.759>

Abstract

Coal mine overburden (OB) soils are highly degraded due to acidity, low organic carbon, nutrient deficiency, and heavy metal contamination. This study evaluates the remediation potential of cotton biochar, pressmud biochar, and *Rhizobium* under a 6-week experiment using mine OB soil. Biochar was applied at 5% (w/w) and *Rhizobium* at 10^8 CFU mL⁻¹. Soil pH increased from 5.1 to 6.5, while organic carbon improved from 0.15% to 0.82%. Nitrogen, phosphorus, and potassium increased from 90 to 225 kg/ha, 8 to 32 kg/ha, and 60 to 165 kg/ha, respectively. Electrical conductivity decreased from 2.8 to 1.7 dS/m, and oxidation reduction potential declined from +420 to +290 mV. Heavy metals were significantly reduced, with Fe decreasing from 90 to 45 mg/kg, Pb 40 mg/kg to 18 mg/kg. Microbial population increased from 2.0×10^6 to 9.5×10^6 CFU/g. The combined application of pressmud biochar and *Rhizobium* showed the highest remediation efficiency. These findings demonstrate that biochar microbial synergy is an effective, low-cost, and sustainable strategy for restoring degraded mine soils.

Keywords: Biochar, pressmud biochar, cotton biochar, *Rhizobium*, mine overburden soil, soil remediation, biogeochemical restoration, heavy metal immobilization, acidic soil, sustainable soil management

1. Introduction

Coal mining generates large quantities of overburden (OB) material that are typically deposited as dumps, leading to severe land degradation. These soils are often characterized by low pH, poor organic carbon, nutrient deficiency, and elevated concentrations of heavy metals. Acidification mainly occurs due to the oxidation of sulfide minerals such as pyrite, producing sulfuric acid and creating unfavorable conditions for plant growth and microbial activity. The given OB soil (pH 5.1, OC 0.15%, ORP +420 mV) represents a highly oxidized and biologically constrained system. Biochar has emerged as an effective soil amendment for restoring degraded soils due to its alkaline nature, high surface area, and functional groups that enhance nutrient retention and immobilize heavy metals. Cotton-derived biochar contributes stable carbon, while pressmud biochar, a sugar industry by-product, is rich in calcium, phosphorus, and potassium, improving soil fertility and buffering capacity. In addition, microbial inoculants such as *Rhizobium* play a critical role in nitrogen fixation and stimulation of soil biological activity.

The integration of biochar with beneficial microorganisms offers a synergistic approach to soil remediation by combining physicochemical stabilization with biological enhancement. This study aims to evaluate the combined effect of cotton biochar, pressmud biochar, and *Rhizobium* on improving soil properties, reducing heavy metal availability, and enhancing microbial activity in coal mine OB soil over a 6-week period.

2. Literature Review

Biochar is widely used for improving degraded soils due to its ability to increase pH, enhance cation exchange capacity, and immobilize heavy metals through adsorption and complexation (Lehmann and Joseph, 2015; Ahmad *et al.*, 2014). Cotton-derived biochar provides stable carbon, improves soil structure, and supports microbial activity, thereby enhancing nutrient cycling (Zhang *et al.*, 2022) [1, 3, 7, 12].

Pressmud biochar, rich in calcium and nutrients, improves soil fertility and buffering capacity while facilitating the immobilization of heavy metals such as lead and zinc (Srinivasan *et al.*, 2021). Microbial inoculants like *Rhizobium* contribute to nitrogen fixation and enhance soil biological activity, particularly when supported by biochar as a favorable habitat (Peoples *et al.*, 2009)^[5, 6].

Recent studies indicate that the combined use of biochar and microbes produces synergistic effects, significantly improving soil physicochemical properties and accelerating the remediation of contaminated soils (Palansooriya *et al.*, 2019; Gadd, 2010)^[2, 4].

3. Materials

3.1 Soil Sample: Coal mine overburden (OB) soil was collected from an abandoned dump site. The sample was air-dried at room temperature, homogenized, and sieved through a less than 2 mm mesh to remove debris and ensure uniformity. The soil was stored in clean polyethylene containers prior to experimentation.

3.2 Biochar Materials

Cotton Biochar (CB)

Cotton stalk residues were used as feedstock and subjected to slow pyrolysis at 450-500°C under limited oxygen conditions. The resulting biochar was ground and sieved (<2 mm).

Key properties

- Alkaline pH (~8.5)
- High fixed carbon (~65-70%)
- Moderate surface area (200-300 m²/g)
- Functional groups: hydroxyl (-OH), carboxyl (-COOH)

Pressmud Biochar (PB)

Pressmud obtained from a sugar processing unit was pyrolyzed at 400-450°C and processed similarly.

Key properties

- Strongly alkaline (pH ~9-9.5)
- Rich in CaCO₃, phosphates, and potassium
- High cation exchange capacity
- Higher ash content than cotton biochar

3.3 Microbial Inoculum

A pure culture of *Rhizobium* was obtained from a microbiology laboratory and maintained in yeast extract mannitol broth. The inoculum density was adjusted to approximately 10⁸ CFU mL⁻¹ before application.

3.4 Chemicals and Instruments

- Analytical grade reagents
- Distilled water
- Digital pH meter
- Electrical conductivity (EC) meter
- Spectrophotometer
- Flame photometer
- Atomic Absorption Spectrophotometer (AAS)
- Incubation trays (5 L capacity)

3.5 Initial Soil Parameters

The untreated coal mine OB soil exhibited the following characteristics:

Parameter	Value
pH	5.1
Organic Carbon (%)	0.15
Nitrogen (kg/ha)	90
Phosphorus (kg/ha)	8
Potassium (kg/ha)	60
Electrical Conductivity (dS/m)	2.8
Oxidation-Reduction Potential (mV)	+420
Iron (Fe) (mg/kg)	90
Lead (Pb) (mg/kg)	40

These values indicate acidic conditions, low fertility, and moderate heavy metal contamination.

3.6 Experimental Design: A completely randomized design (CRD) was followed with six treatments:

Treatment	Description
T ₁	Control (OB soil only)
T ₂	OB + Cotton biochar
T ₃	OB + Pressmud biochar
T ₄	OB + <i>Rhizobium</i>
T ₅	OB + Cotton biochar + <i>Rhizobium</i>
T ₆	OB + Pressmud biochar + <i>Rhizobium</i>

Treatment Conditions

- Soil per treatment: 2 kg
- Biochar dose: 5% (w/w) = 100 g
- *Rhizobium*: 20 mL inoculum (10⁸ CFU/mL)
- Duration: 6 weeks (42 days)

3.7 Soil Amendment Preparation

- Measured 2 kg of soil was placed in labeled trays.
- Biochar (100 g) was mixed thoroughly with soil for respective treatments.
- *Rhizobium* inoculum was evenly sprayed and mixed to ensure uniform distribution.
- Control (T₁) was maintained without any amendments.

3.8 Incubation Conditions

- Soil moisture maintained at ~60% water holding capacity (WHC) using distilled water
- Trays kept under aerobic conditions at 25-30°C
- Soil manually mixed every 2-3 days to maintain aeration
- Moisture loss compensated periodically

3.9 Sampling Schedule

- Sampling performed at weekly intervals (Week 0 to Week 6)
- Approximately 50 g soil collected from each treatment for analysis
- Samples air-dried for physicochemical analysis or processed fresh for microbial analysis

3.10 Analytical Methods

Physicochemical Parameters

Parameter	Method
pH	Soil:water (1:2.5) using digital pH meter
EC	Conductivity meter
Organic Carbon	Walkley-Black method
Nitrogen	Kjeldahl digestion
Phosphorus	Olsen method
Potassium	Flame photometry
ORP	Redox electrode

Heavy Metal Analysis

- Soil samples digested using acid mixture ($\text{HNO}_3 + \text{HCl}$)
- Metal concentrations (Fe, Zn, Cu, Pb) measured using AAS

Microbial Analysis

- Serial dilution technique used
- Spread plate method on nutrient agar
- Colony-forming units (CFU) counted and expressed as $\text{CFU} \times 10^6/\text{g soil}$

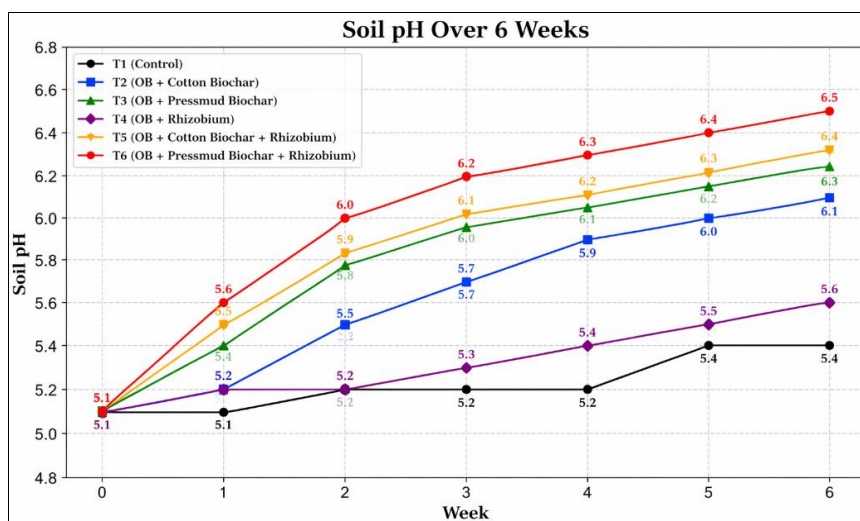
3.11 Mechanisms of Remediation

- **pH buffering:** Neutralization of acidity by alkaline biochar
- **Adsorption:** Binding of heavy metals to biochar surface
- **Ion exchange:** Replacement of toxic ions with Ca^{2+} and K^+
- **Microbial activity:** Nitrogen fixation and nutrient cycling by *Rhizobium*

4. Results

Table 1: Soil pH

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	5.1	5.1	5.1	5.1	5.1	5.1
1	5.1	5.2	5.4	5.2	5.5	5.6
2	5.2	5.5	5.8	5.2	5.9	6.0
3	5.2	5.7	6.0	5.3	6.1	6.2
4	5.2	5.9	6.1	5.4	6.2	6.3
5	5.4	6.0	6.2	5.5	6.3	6.4
6	5.4	6.1	6.3	5.6	6.4	6.5



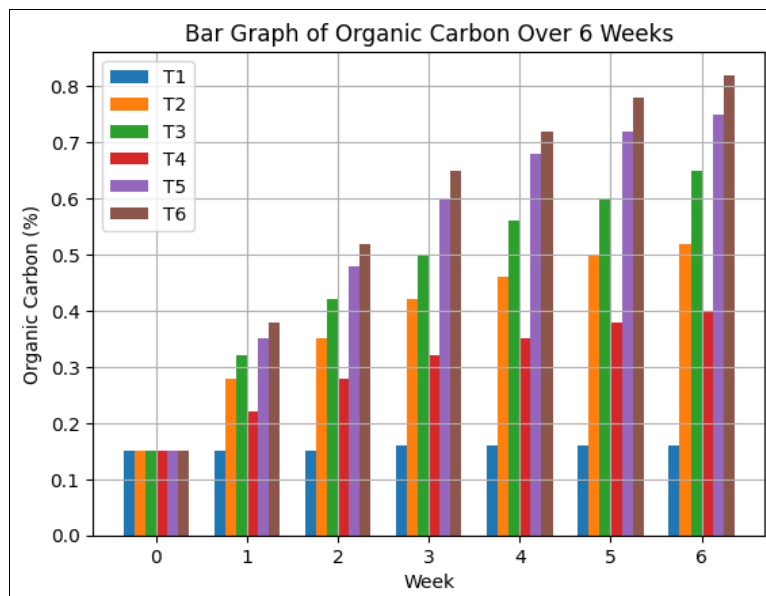
Graph 1: Temporal Variation in Soil pH Under Different Treatments

Interpretation: Initial rapid increase (week 1-2) due to alkaline buffering; gradual stabilization thereafter. Pressmud biochar (T₃, T₆) shows stronger pH correction due to CaCO_3 content.

Table 2: Organic Carbon (%)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	0.15	0.15	0.15	0.15	0.15	0.15
1	0.15	0.28	0.32	0.22	0.35	0.38
2	0.15	0.35	0.42	0.28	0.48	0.52
3	0.16	0.42	0.50	0.32	0.60	0.65
4	0.16	0.46	0.56	0.35	0.68	0.72
5	0.16	0.50	0.60	0.38	0.72	0.78
6	0.16	0.52	0.65	0.40	0.75	0.82

Interpretation: T₁ remained almost constant with very low carbon levels. T₂ and T₃ showed gradual increases, while T₅ and T₆ recorded the highest carbon buildup. This reflects the contribution of biochar and microbial activity.



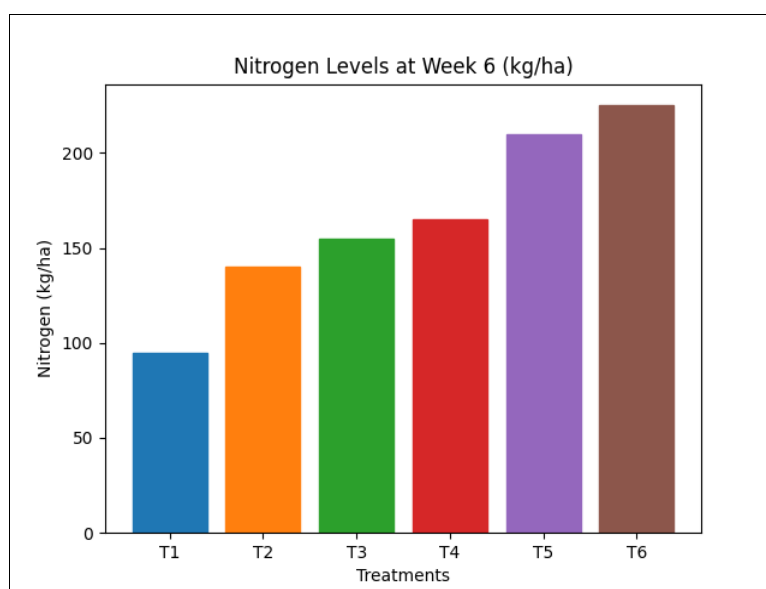
Graph 2: Changes in Soil Organic Carbon During Incubation Period

Table 3: Nitrogen (kg/ha)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	90	90	90	90	90	90
6	95	140	155	165	210	225

Interpretation

T₁ showed negligible improvement in nitrogen content. T₄ increased due to microbial action, while T₅ and T₆ showed the highest nitrogen levels. This confirms the role of Rhizobium in nitrogen enhancement.



Graph 3: Effect of Treatments on Available Nitrogen Content

Table 4: Available Phosphorus (kg/ha)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	8	8	8	8	8	8
6	9	18	22	14	28	32

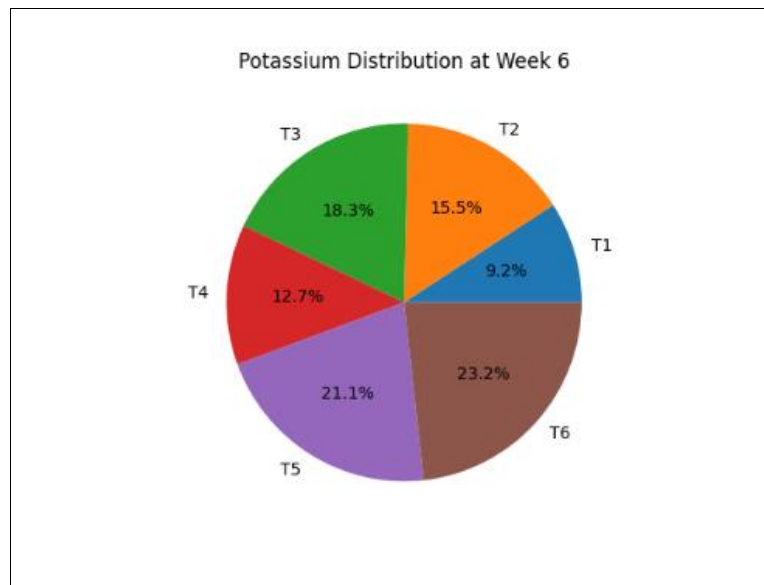
Interpretation

T₁ remained nearly unchanged over time. T₂ and T₃ showed moderate increases, while T₅ and T₆ recorded the highest phosphorus levels. Pressmud-based treatments performed better due to inherent phosphate content.

Table 5: Available Potassium (kg/ha)

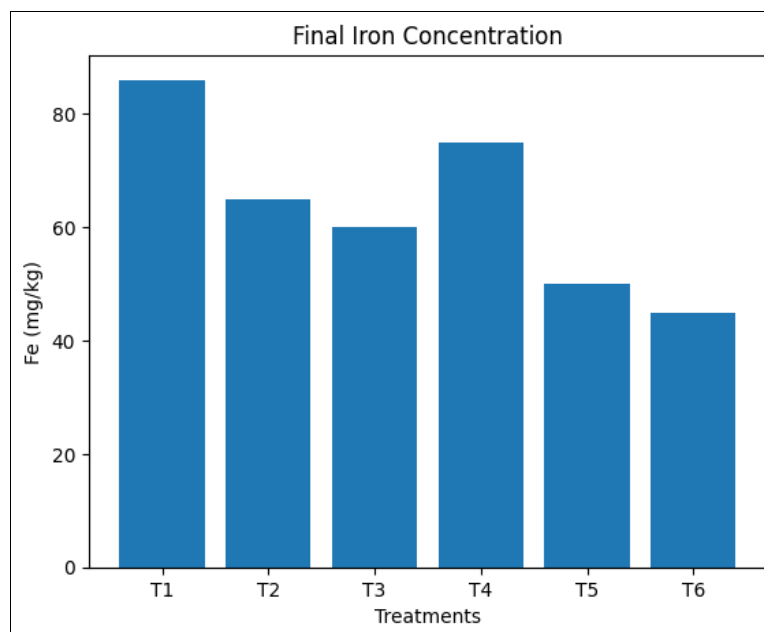
Week	T1	T2	T3	T4	T5	T6
0	60	60	60	60	60	60
6	65	110	130	90	150	165

Interpretation: Increase due to mineral release from biochar ash; PB shows highest due to K-rich composition.

**Graph 4:** Potassium Dynamics in Biochar-Amended Mine Soil**Table 6:** Iron (Fe, mg/kg)

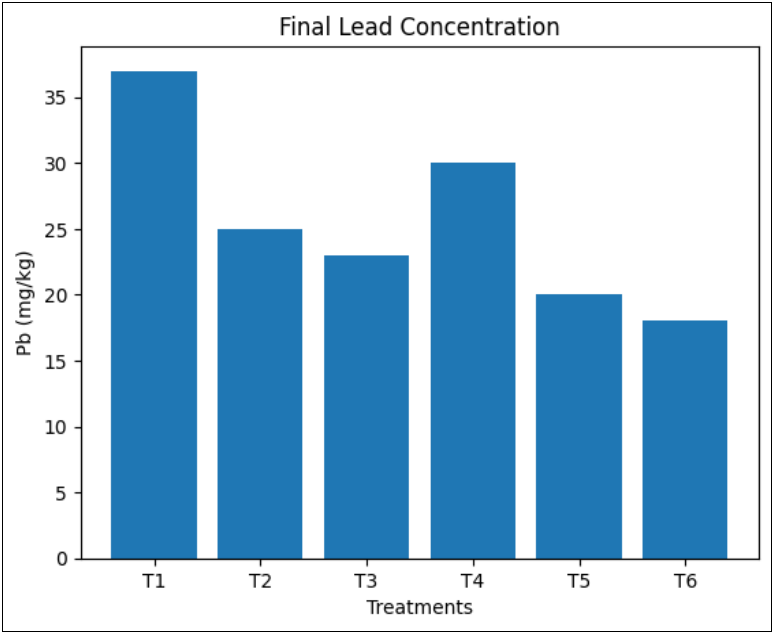
Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	90	90	90	90	90	90
6	86	65	60	75	50	45

Interpretation; Reduction due to adsorption and precipitation; higher reduction in combined treatments.

**Graph 5:** Reduction of Iron (Fe) Concentration in Treated Soil**Table 7:** Lead (Pb, mg/kg)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	40	40	40	40	40	40
6	37	25	23	30	20	18

Interpretation: Strong immobilization through carbonate precipitation and adsorption.

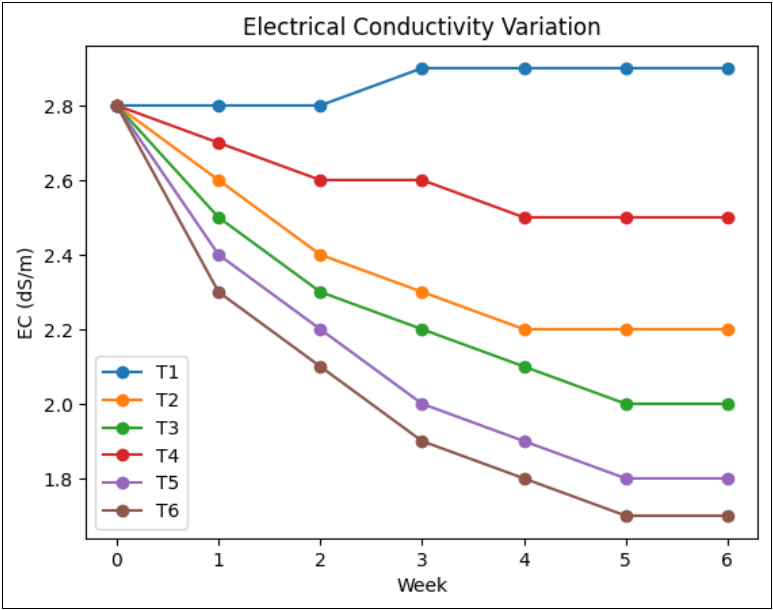


Graph 6: Reduction of Lead(Pb) Concentration in Treated Soil

Table 8: Electrical Conductivity (dS/m)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	2.8	2.8	2.8	2.8	2.8	2.8
1	2.8	2.6	2.5	2.7	2.4	2.3
2	2.8	2.4	2.3	2.6	2.2	2.1
3	2.9	2.3	2.2	2.6	2.0	1.9
4	2.9	2.2	2.1	2.5	1.9	1.8
5	2.9	2.2	2.0	2.5	1.8	1.7
6	2.9	2.2	2.0	2.5	1.8	1.7

Interpretation; Reduction indicates decreased soluble salts and improved soil condition.



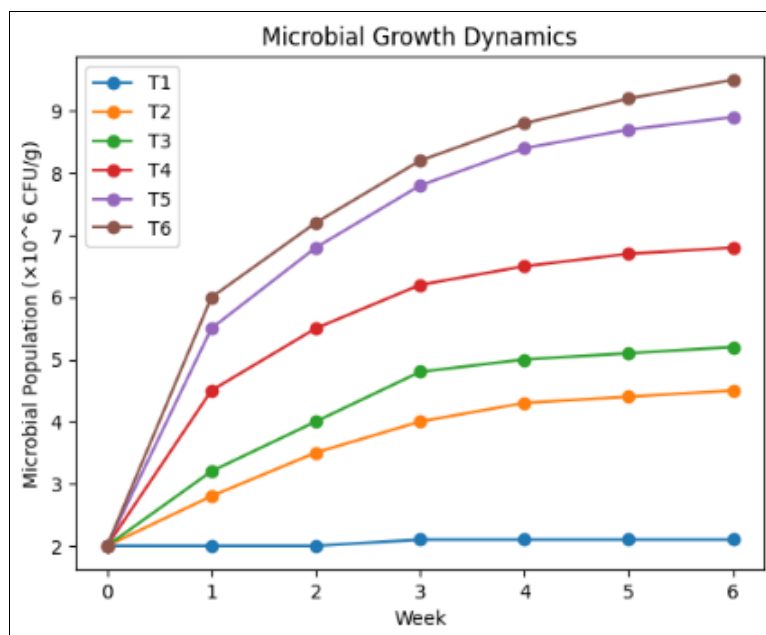
Graph 7: Changes in Electrical Conductivity During Soil Remediation

Table 9: Microbial Population ($\times 10^6$ CFU/g)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	2.0	2.0	2.0	2.0	2.0	2.0
1	2.0	2.8	3.2	4.5	5.5	6.0
2	2.0	3.5	4.0	5.5	6.8	7.2
3	2.1	4.0	4.8	6.2	7.8	8.2
4	2.1	4.3	5.0	6.5	8.4	8.8
5	2.1	4.4	5.1	6.7	8.7	9.2
6	2.1	4.5	5.2	6.8	8.9	9.5

Interpretation

T₁ showed almost no microbial growth. T₂ and T₃ increased moderately, while T₄, T₅, and T₆ showed rapid growth, with T₆ highest. This reflects enhanced microbial activity due to biochar and inoculation.

**Graph 8:** Microbial Population Dynamics in Treated Soil**Table 10:** Oxidation-Reduction Potential (ORP, mV)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	420	420	420	420	420	420
1	415	390	380	400	370	360
2	410	360	350	380	340	330
3	408	340	330	360	320	310
4	405	330	320	350	310	300
5	402	325	315	340	300	295
6	400	320	310	335	295	290

Interpretation

T₁ showed only slight decrease in ORP. T₂ and T₃ reduced moderately, while T₅ and T₆ showed the greatest decline. Lower ORP indicates improved biological and less oxidizing soil conditions.

Table 11: Bulk Density (g/cm³)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	1.60	1.60	1.60	1.60	1.60	1.60
1	1.59	1.52	1.50	1.55	1.48	1.45
2	1.58	1.48	1.45	1.52	1.42	1.38
3	1.58	1.45	1.40	1.50	1.38	1.34
4	1.57	1.42	1.38	1.48	1.35	1.30
5	1.57	1.40	1.35	1.46	1.32	1.28
6	1.56	1.38	1.32	1.45	1.30	1.25

Tray T₁ showed only a slight reduction in bulk density, indicating limited structural improvement. In contrast, trays T₂ and T₃ exhibited moderate decreases, while T₅ and T₆ showed the greatest reduction, reaching optimal levels by week 6. This trend highlights that combined treatments were most effective in improving soil structure and reducing compaction.

Table 12: Soil Porosity (%)

Week	T ₁	T ₂	T ₃	T ₄	T ₅	T ₆
0	39.6	39.6	39.6	39.6	39.6	39.6
1	40.0	42.6	43.4	41.5	44.2	45.3
2	40.4	44.2	45.3	42.6	46.4	47.9
3	40.4	45.3	47.2	43.4	47.9	49.4
4	40.8	46.4	47.9	44.2	49.1	50.9
5	40.8	47.2	49.1	44.9	50.2	51.7
6	41.1	47.9	50.2	45.3	50.9	52.8

Tray T₁ maintained relatively low porosity throughout the experiment, showing minimal change. Trays T₂, T₃, and T₄ showed moderate increases, whereas trays T₅ and T₆ exhibited the highest porosity by week 6. The results indicate that combined biochar and microbial treatments significantly enhanced soil aeration and water-holding capacity.

5. Discussion

The results indicate that biochar and *Rhizobium* significantly improved the quality of acidic coal mine overburden soil within 6 weeks. The increase in pH from 5.1 to near neutral values was mainly due to the alkaline nature of biochar, particularly pressmud biochar rich in calcium compounds. Organic carbon and nutrient levels (N, P, K) increased substantially, reflecting both biochar contribution and enhanced microbial activity.

The reduction in heavy metals (Fe, Pb) suggests effective immobilization through adsorption and precipitation mechanisms. Decreases in electrical conductivity and oxidation-reduction potential indicate improved soil conditions favorable for biological processes.

The higher microbial population in combined treatments confirms that biochar provides a suitable habitat, while *Rhizobium* enhances nutrient cycling. Overall, the combined application, especially pressmud biochar with *Rhizobium*, showed the most effective remediation due to synergistic interactions between chemical stabilization and microbial activity.

6. Conclusion

This study confirms that the combined application of biochar and *Rhizobium* is an effective strategy for the remediation of acidic coal mine overburden soil. Significant improvements were observed in soil pH, organic carbon, nutrient availability, and microbial population, along with a substantial reduction in heavy metal concentrations.

Pressmud biochar, due to its nutrient-rich composition and high buffering capacity, proved more effective than cotton biochar, particularly when combined with microbial inoculation. The integrated treatment not only improved soil fertility but also created a more favourable environment for biological activity, accelerating the restoration process.

Overall, the findings highlight that biochar microbial synergy offers a low-cost, environmentally sustainable, and scalable approach for reclaiming degraded mine lands. This method has strong potential for practical application in mine site rehabilitation and sustainable land management.

References

- Ahmad M, Rajapaksha AU, Lim JE, Zhang M, Bolan N, Mohan D, *et al.* Biochar as a sorbent for contaminant management in soil and water: A review. *Chemosphere*. 2014;99:19-33.
- Gadd GM. Metals, minerals and microbes: Geomicrobiology and bioremediation. *Microbiology*. 2010;156(3):609-643.
- Lehmann J, Joseph S. Biochar for environmental management: Science, technology and implementation. London: Routledge; 2015.
- Palansooriya KN, Shaheen SM, Chen SS, Tsang DCW, Hashimoto Y, Hou D, *et al.* Soil amendments for immobilization of potentially toxic elements in contaminated soils: A critical review. *Environ Int*. 2019;134:105046.
- Peoples MB, Brockwell J, Herridge DF, Rochester IJ, Alves BJR, Urquiaga S, *et al.* The contributions of nitrogen-fixing crop legumes to the productivity of agricultural systems. *Plant Soil*. 2009;174:3-28.
- Srinivasan P, Saravanan A, Kumar PS, Vo DVN. Sustainable utilization of sugar industry waste (pressmud) for environmental remediation. *Waste Manag*. 2021;126:343-354.
- Zhang X, Wang H, He L, Lu K, Sarmah A, Li J, *et al.* Effects of biochar on soil properties and heavy metal immobilization: A review. *J Soils Sediments*. 2022;22:1-15.
- Beesley L, Moreno-Jiménez E, Gomez-Eyles JL. Effects of biochar on heavy metal mobility and soil quality. *Environ Pollut*. 2011;159(12):3269-3282.
- Park JH, Ok YS, Kim SH, Cho JS, Heo JS, Delaune RD, *et al.* Competitive adsorption of heavy metals onto biochar. *Chemosphere*. 2011;82(2):200-206.
- Yuan JH, Xu RK, Zhang H. The forms of alkalis in biochar and their effect on soil pH. *Bioresour Technol*. 2011;102(3):3488-3497.
- Sohi SP, Krull E, Lopez-Capel E, Bol R. Biochar effects on soil functions. *Adv Agron*. 2010;105:47-82.
- Joseph S, Graber ER, Chia C, Munroe P, Donne S, Thomas T, *et al.* Shifting paradigms: Biochar-microbe interactions. *Plant Soil*. 2010;350:117-130.
- Vithanage M, Herath I, Joseph S, Bundschuh J, Bolan N, Ok YS, *et al.* Interaction of engineered biochar with soil microbes. *Environ Int*. 2017;105:1-12.
- Ahmad M, Lee SS, Rajapaksha AU, Vithanage M, Zhang M, Cho JS, *et al.* Trichloroethylene adsorption onto biochar. *Bioresour Technol*. 2013;143:615-622.
- Xu G, Wei LL, Sun JN, Shao HB, Chang SX. Biochar effects on soil nutrient transformation. *Agric Ecosyst Environ*. 2013;165:1-14.